AMENDMENTS TO THE SPECIFICATION

Page 2, before line 1, insert the following replacement paragraph:

CROSS-REFERENCE TO RELATED APPLICATIONS

This application is a division of U.S. patent application Serial No. 09/218,176 filed December 22, 1998, now U.S. Patent 6,171,584, which in turn is a continuation of U.S. patent application Serial No. 08/679,048 filed July 12, 1996, which is a continuation-in-part of U.S. patent application Serial No. 08/482,577, filed ~~July 6~~ June 7, 1995, now U.S. patent number 5,807,713. The disclosures of the prior applications are incorporated by reference herein in their entirety.

Page 5, fourth full paragraph onto page 6, insert the following replacement paragraph:

Figure 1 shows a comparison of the amino acid sequence of human MP121 (SEQ ID NO:7) with some members of the TGF-β family (inhibin α and β chains) (SEQ ID NOS:8-10) starting at the first of the seven conserved cysteine residues. * denotes that the amino acid is the same in all compared proteins; + denotes that the amino acid corresponds in at least one of the proteins compared to ~~humna~~ human MP121.

Page 6, second full paragraph, insert the following replacement paragraph:

Figure 3 shows a diagram of a Western blot using chicken antibodies against human MP121. Lane 1 shows *E. coli* cells transformed with pBP4MP121His under reducing conditions (1% β-mercaptoethanol). Lane 2 shows cell culture supernatant of NIH-3T3 cells after infection with recombinant viruses (with inserted MP121 cDNA) under reducing conditions (1% β-mercaptoethanol). Lane 3 shows cell culture supernatant of NIH-3T3 cells after infection with recombinant viruses (with inserted MP121 cDNA) under non-reducing conditions. Lane M shows

prestained protein molecular weight markers having the stated apparent molecular weights (Gibco BRL #26041-020).

Page 6, third full paragraph, insert the following replacement paragraph:

Figure 4 shows the expression of MP121 compared to activin β_A and β_B in various mouse tissues and is an autoradiogram after gel analysis of an RNase protection assay using specific probes against activin βA (βA), activin βB (βB), MP121 and against GAPDH for the control. Total RNA was tested which has been isolated from various mouse tissues (Lane 1: brain; Lane 2: heart, Lane 3: kidney, Lane 4: liver, Lane 5: lung, Lane 6: muscle, Lane 9: ovary, Lane 10: spleen, Lane 11: testes) from embryonic stem cells (Lane 12: CJ7) and from yeast (Lane 13) as a control. No RNA was used in Lane 14 as a control. The unprotected antisense RNA probes used for the hybridization are applied in lanes 8 and 15 and the expected fragment size is indicated in brackets in the right margin. The bands of the protected fragments are labeled in the left margin. PBR322 restricted with ~~Map~~ Msp 1 (Biolabs #303) and end-labeled with γ-32p-ATP (Amersham) was used as the marker (Lane 7).

Page 24, line 12 to page 25, line 30, insert the following replacement paragraph:

As an example, the mature part of human MP121 (amino acid 237 to 352 in SEQ ID NO:2) with an additional 13 amino acids, including six histidines at the N-terminus, (MHHHHHHKLEFAM) (SEQ ID NO:40) was expressed in the prokaryotic vector pBP4. This vector is a pBR322 derivative having ~~tetracyclin~~ tetracycline resistance which, in addition, contains the T7 promoter from the pBluescript II SK plasmid (Stratagene). Furthermore, the vector contains a ribosomal binding site following the T7 promoter and a start codon followed by six codons for histidine. A terminator (TØ) follows after several single restriction cleavage sites such as EcoRI, XhoI, SmaI and ApaI, for the insertion of inserts as well as stop codons in all three reading frames. In order to obtain the cDNA for the mature part of MP121, PCR was carried out on the plasmid

SK121L9.1 (DSM ~~dispositary~~ depository number: 9177) using the two oligonuleotides GAATTCGCCATGGGCATCGACTGCCAAGGAGG (SEQ ID NO:41) and CCGCTCGAGAAGCTTCAACTGCACCCACAGGC (SEQ ID NO:42). Both oligonucleotides contain additional restriction cleavage sites at their ends (EcoRI and NcoI or XhoI and HindIII). In an intermediate step the resulting 377 bp fragment was cloned with blunt ends into the pBluescript II SK vector (~~Stragene~~ Stratagene) that had been cleaved with EcoRV. One clone in the orientation of the 5' end of MP121 towards the T7 promoter was cleaved with EcoRI and the resulting insert (0.38 kb) was cloned into the pBP4 vector that had also been cleaved with EcoRI. The correct orientation of the insert in the resulting plasmid pBP4MP121His was established by restriction analysis and sequencing. The plasmid pBR4MP121His was deposited on 30.1.1995 at the DSM (~~depositary~~ depository number: 9704). The expression of MP121 protein can be achieved by simultaneously providing T7 RNA polymerase. T7 RNA polymerase can be provided by various methods such as, e.g., by a second plasmid with a gene for T7 RNA polymerase or by infection with phages which code for T7 RNA polymerase or also by special bacterial strains which have integrated the gene for T7 RNA polymerase. The mature MP121 protein with a His-tag (MP121His) is produced in inclusion bodies by using the bacterial strain BL21 (DE3)pLysS (Novagen, #69451-1) and inducing the T7 RNA polymerase expression with IPTG according to the manufacturer's instructions. In SDS polyacrylamide gels (15%) the protein exhibits an apparent molecular weight of nearly 16 kD (theoretical molecular weight: 14.2 kD) as is shown representatively in the Western blot of Fig. 3. The bacteria transformed with pBP4 as controls do not show any staining of specific bands. Due to the His-tag this protein can be purified on nickel-chelating agent columns as described, for example, by Hochuli et al., (Bio/Technology, Vol. 6, 1321-1325 (1988)). An additional purification is possible by means of reversed phase HPLC. A reversed phase column (Nucleosil 300-7C4 from Macherey-Nagel, Type 715023) was used with a flow-rate of 2 ml/min and an acetonitrile gradient in 0.1% TFA of 0 to 90% within 100 minutes. MP121His elutes under these conditions after ca. 40% acetonitrile.

Page 28, lines 19-33, insert the following replacement paragraph:

PCR reactions and intermediate cloning were necessary in order to shorten the 5' and 3' untranslated regions of the initial plasmid SK121L9.1 (DSM, depositary number: 9177) and to insert single restriction cleavage sites at the ends. All PCR reactions were carried out using the plasmid SK121L9.1 (DSM depositary number: 9177). In order to shorten the 5' untranslated end, the primer CCCGGATCCGCTAGCACCATGACCTCCTCATTGCTTCTG (SEQ ID NO:44) with an inserted BamHI and NheI restriction cleavage site was used in a PCR with an internal primer (CCCTGTTGTCCTCTAGAAGTG) (SEQ ID NO:45). In an intermediate step, the fragment obtained was cloned into Bluescript SK (Stratagene), sequenced and checked for concurrence with the sequence shown in SEQ ID NO:1. The SphI/EcoRI fragment (0.22 kb) from the plasmid ~~pBR4MP121His~~ pBP4MP121His was used to shorten the 3' untranslated end.

Page 32, lines 7-22, insert the following replacement paragraph:

Total RNA from various tissues (brain, heart, kidney, liver, lung, spleen, muscle, ovary, testes) was isolated according to standard methods from six-week-old mice as well as from embryonic stem cells. 10 µg total RNA was used in each case in a RNAse protection assay (RPA) from Ambion (RPA II kit, #1410) according to the manufacturer's instructions. In order to obtain specific probes for activin β_A and activin β_B the genomic DNA from the mouse (129Sv) was amplified from the mature part of the proteins using corresponding specific primers. In order to facilitate cloning, EcoRI and/or BamHI or HindIII restriction cleavage sites were introduced, respectively, at the ends of the primers. In the case of activin β_A the primers were derived from mRNA from rats (GenBank Accession #M37482):

~~GGATCCGAATTCGGCTTGGAGTGTATGGCAAGG~~

GGATCCGAATTCGGCTTGGAGTGTGATGGCAAGG (SEQ ID NO:46)

and GGATCCGAATTCCTCTGGGACCTGGCAACTCTAG (SEQ ID NO:47).

AMENDMENTS TO THE CLAIMS

This listing of claims will replace all prior versions, and listings, of claims in the application.

Listing of Claims

Claims 1-23 (canceled).

Claim 24 (currently amended). An isolated protein of the TGF-β family which has mitogenic and/or differentiation-inductive activity and is coded by a DNA molecule selected from the group consisting of

(a) a DNA molecule comprising the nucleotide sequence shown in the SEQ ID No:1, or the following fragments: nucleotides 128-1183 of SEQ ID NO:1, nucleotides 836-1183 of SEQ ID NO:1, nucleotides 128-835 of SEQ ID NO:1, and nucleotides 866-1183 of SEQ ID NO:1;

(b) a DNA molecule comprising the nucleotide sequence shown in the SEQ ID No:3, or the following fragments: nucleotides 131-1186 of SEQ ID NO:3, nucleotides 839-1186 of SEQ ID NO:3, nucleotides 131-838 of SEQ ID NO:3, and nucleotides 869-1186 of SEQ ID NO:3;

(c) a DNA molecule encoding the amino acid sequence encoded by (a) or (b); and

(d) a DNA molecule comprising a nucleotide sequence which hybridizes with one of the DNA molecules from (a) and (b) under stringent hybridization conditions in 6x SSC at 62-66° C followed by one hour wash with 0.6x SSC and 0.1% SDS at 62-66° C and which encodes a protein comprising an amino acid sequence containing seven conserved cysteine residues, said seven conserved cysteines corresponding to cysteine residues at positions 247, 276, 280, 316, 317, 349 and 351 in SEQ ID NO:2.

Claim 25 (currently amended). The protein according to claim 24, wherein said protein has an amino acid sequence selected from the group consisting of SEQ ID NO:2; SEQ ID NO:4; a part of SEQ ID NO:2 corresponding to ~~the~~ a mature protein, wherein said mature protein ~~which~~ starts

with one of amino acids 217-240 and ends with amino acid 352 of SEQ ID NO:2; and a part of SEQ ID NO:4 corresponding to ~~the~~ a mature protein, wherein said mature protein ~~which~~ starts with one of amino acids 217-240 and ends with amino acid 352 of SEQ ID NO:4~~; and sequences containing conservative substitutions of the amino acids shown in SEQ ID NO:2, SEQ ID NO:4 or the said parts thereof~~.

Claim 26 (previously presented). A heterodimeric protein comprising a monomer of the protein of claim 24 and a monomer of another protein from the TGF-β family.

Claim 27 (currently amended). A pharmaceutical composition comprising the protein of claim 24 and ~~pharmaceutical~~ pharmaceutically acceptable carrier or auxiliary substances, diluents or fillers.

Claim 28 (previously presented). A pharmaceutical composition comprising the protein of claim 26 and pharmaceutically acceptable carrier or auxiliary substances, diluents or fillers.

Claim 29 (currently amended). The protein according to claim 24, wherein said protein has an amino acid sequence selected from the group consisting of SEQ ID NO:2, SEQ ID NO:4, the part of SEQ ID NO:2 corresponding to the mature protein which ~~includes~~ comprises at least the region of ~~the~~ seven cysteine residues, said region comprising amino acid residues 247-351 of SEQ ID NO:2, and the part of SEQ ID NO:4 corresponding to ~~the~~ a mature protein which ~~includes~~ comprises at least the region of ~~the~~ seven cysteine residues, said region comprising amino acid residues 247-351 of SEQ ID NO:4 ~~and sequences containing conservative substitutions of the amino acids shown in SEQ ID NO:2, SEQ ID NO:4 or the mature parts thereof~~.

Claim 30 (currently amended). The heterodimeric protein of claim 26, wherein the other member of the TGF-β family is activin/inhibin or a ~~BMP~~ bone morphogenetic protein.

Claim 31 (previously presented). A homodimeric protein comprising two monomers of the protein of claim 24.

Claim 32 (previously presented). A monomeric protein comprising the protein of claim 24.

Claim 33 (previously presented). A pharmaceutical composition comprising the protein of claim 31 and pharmaceutically acceptable carrier or auxiliary substances, diluents or fillers.

Claim 34 (previously presented). A pharmaceutical composition comprising the protein of claim 32 and pharmaceutically acceptable carrier or auxiliary substances, diluents or fillers.

Claim 35 (new). The protein according to claim 25, wherein said protein has an amino acid sequence selected from the group consisting of a part of SEQ ID NO:2 corresponding to a mature protein, wherein said mature protein starts with amino acid 236 and ends with amino acid 352 of SEQ ID NO:2, a part of SEQ ID NO:2 corresponding to a mature protein, wherein said mature protein starts with amino acid 237 and ends with amino acid 352 of SEQ ID NO:2, a part of SEQ ID NO:4 corresponding to a mature protein, wherein said mature protein starts with amino acid 236 and ends with amino acid 352 of SEQ ID NO:4 and a part of SEQ ID NO:4 corresponding to a mature protein, wherein said mature protein starts with amino acid 237 and ends with amino acid 352 of SEQ ID NO:4.

Claim 36 (new). A pharmaceutical composition comprising the protein of claim 35 and pharmaceutically acceptable carrier or auxiliary substances, diluents or fillers.

Claim 37 (new). A heterodimeric protein comprising a monomer of the protein of claim 35 and a monomer of another protein from the TGF-β family.

Claim 38 (new). A pharmaceutical composition comprising the protein of claim 36 and pharmaceutically acceptable carrier or auxiliary substances, diluents or fillers.

Claim 39 (new). The heterodimeric protein of claim 36, wherein the other member of the TGF-β family is activin/inhibin or a bone morphogenetic protein.

Claim 40 (new). A homodimeric protein comprising two monomers of the protein of claim 35.

Claim 41 (new). A pharmaceutical composition comprising the protein of claim 40 and pharmaceutically acceptable carrier or auxiliary substances, diluents or fillers.

Claim 42 (new). A monomeric protein comprising the protein of claim 35.

Claim 43 (new). A pharmaceutical composition comprising the protein of claim 42 and pharmaceutically acceptable carrier or auxiliary substances, diluents or fillers.

REMARKS

The amendments to the specification are made to correct minor typographical errors, and do not represent the addition of new matter.

Claim 24 has been amended to insert the sequence identifier and to specify the position of the seven conserved cysteines as shown in Figure 1 with reference to the sequence set forth in SEQ ID NO:2 or 4. Claim 25 has been amended to more clearly define the nature of the mature proteins, i.e., a protein starting with one of amino acids 217-240 and ending with amino acid 352. Claim 25 has further been amended to delete reference to proteins having conservative amino acid substitutions. Claim 27 has been amended to correct a typographical error. Claim 29 has been amended to more clearly specify the region of the proteins containing the seven conserved cysteine residues as shown in Figure 1 with reference to the sequence set forth in SEQ ID NO:2. Claim 29 has further been amended to delete reference to proteins having conservative amino acid substitutions. Claim 30 has been amended to set forth the term for the abbreviation BMP.

New claims 35-43 have been added to claim subject matter in which the protein is a part of the mature protein of SEQ ID NOs:2 and 4. Claim 35 is directed to the mature protein which starts with amino acid residue 236 or 237 and ends with amino acid residue 352 of SEQ ID NOs:2 and 4. Support for this amendment can be found at page 4, 4th line from the bottom - page 5, line 23. Claims 36-43 are similar to claims 26-28 and 30-34 with dependency on claim 35.

It is submitted that none of these amendments constitute new matter, and their entry is requested.

In reviewing the filing receipt for correctness, Applicants have discovered an error which originated in the Declaration and Power of Attorney which was prepared for the parent application. This Declaration and Power of Attorney incorrectly identified one of the parent applications as U.S. Serial No. 08/482,557. The correct parent application should be U.S. Serial No. 08/482,577 as set forth in the first paragraph of the application as amended in the previous amendment. Applicants submitted a Substitute Declaration to indicate the correct application numbers on 8 October 2003.

Claims 24-43 are pending in the application.

The Examiner has maintained the rejection of claims 24-28 and 29-34 under 35 U.S.C. § 112, first paragraph, for lack of written description, and apparently for lack of enablement in light of her citation of the *In re Wands* case and undue experimentation, both of which relate to enablement and not written description. It is submitted that the amended claims obviate this rejection to the extent that it may have been properly made.

Specifically, in the written description aspect of this rejection, the Examiner focuses on the part of the specified sequences which correspond to the mature protein. As made clear in claim 25, the mature protein starts with one of amino acids 217 through 240 and ends with amino acid 352. Thus, the mature protein specifically begins with residue 217, or residue 218 or residue 219, etc., up to residue 240. The fact that the mature peptide could begin with any one of the residues 217-240 is fully described in the specification at page 4, last line - page 5, line 2 and page 5, lines 13-17. The relevant portions of this part of the specification state:

> The start of the mature protein is preferably in the region of amino acids 217-240, particularly preferably at amino acid 236 or 237 and most preferably at amino acid 237.
>
> ...
>
> The mature protein begins in the region of amino acids 217-240 in analogy with the human MP121 of SEQ ID NO:2. It is most preferred when the mature protein starts at amino acid 237 ... In the case of MP121 from the mouse it is also conceivable that the beginning of the mature protein is at least sometimes amino acid 236.

This description in the specification clearly shows to a skilled artisan that Applicants were in possession of the claimed invention. The Examiner also makes comments concerning the terms "mature protein" and "essentially." However, it is submitted that these comments are not appropriate, since the claim specifically defines the mature protein by the starting and ending residues and the word "essentially" is not used in the claims.

In the enablement aspect of this rejection, the Examiner raised the issue of the location of the cysteine residues with respect to claim 24. Claim 24 has been amended to specify the location of the seven conserved cysteines with respect to SEQ ID NO:2. It is submitted that the amendment of claim 24 obviates this aspect of the rejection. Also, as detailed in the previous amendment, the

proteins of the claims all share the following attributes: (1) they have mitogenic and/or differentiation-inductive activity; (2) they are encoded by the nucleic acids recited in parts (a) - (c) or by nucleic acids which hybridize under stringent hybridization conditions with the nucleic acids recited in parts (a) and (b); and (3) they all contain seven conserved cysteine residues for which their location or the region containing them are specified in the claims. The hybridization is an important attribute which cannot be overlooked, as is the attribute of seven conserved cysteine residues and their position with respect to the sequence identifiers set forth in the claims. The claims do not encompass any proteins which do not contain seven conserved cysteine residues and which are encoded by nucleic acids which do not hybridize to the nucleic acids of parts (a) or (b) under stringent conditions. Applicants note that the language of claim 24 has been drafted in a manner to be consistent with the language of claim 1 of the parent application Serial No. 09/218,176, which the present Examiner allowed and which has matured into U.S. Patent No. 6,171,584.

In addition to the enablement aspect of this rejection, the Examiner also raised the issue of whether protein sequences containing conservative amino acid substitutions were enabled. Claims 25 and 29 have been amended to delete the objected language. It is submitted that the amendment of claims 25 and 29 obviates this aspect of the rejection.

Furthermore, in *In re Wright*, 27 USPQ 2d, 1510 (Fed. Cir. 1993), the Federal Circuit made it clear that the PTO has the burden of providing a reasonable explanation of why the specification does not enable. There must be some reason to doubt the objective truth of the specification statements. *In re Marzocchi*, 169 USPQ 367 (CCPA 1973). Applicants submit that the Examiner has not provided acceptable evidence which is inconsistent with the objective enablement of the specification. Since the Examiner has not presented any scientific evidence or reasons to doubt the objective enablement of the specification with respect to the now claimed proteins, it is submitted that a proper case for lack of compliance with the enablement provision of 35 U.S.C. §112, first paragraph, has not been established. In addition, the mere citation of the *Wands* case and the listing of factors to be considered with no analysis of the factors does not comply with the Patent Office's guidelines for such rejections.

In view of the above remarks and the amendments to the claims, it is submitted that claims 24-28 and 29-34 fully comply with the requirements of 35 U.S.C. § 112, first paragraph. Withdrawal of this rejection is requested.

Claims 24-34 were rejected under 35 U.S.C. § 112, second paragraph, as indefinite. The claims have been amended to specify the sequence identifier, provide proper antecedent basis, specify the position or the region of the seven conserved cysteines and to set forth the term for BMP. It is submitted that these amendments obviate this rejection, and its withdrawal is requested.

In view of the above amendments and remarks, it is believed that the claims satisfy the requirements of the patent statutes and are patentable over the prior art. Reconsideration of the instant application and early notice of allowance are requested. The Examiner is invited to telephone the undersigned if it is deemed to expedite allowance of the application.

RESPECTFULLY SUBMITTED,					
Name and Reg. Number	Jeffrey L. Ihnen, Registration No. 28,957				
Signature			*Date*	29 October 2003	
Address	Rothwell, Figg, Ernst & Manbeck 1425 K Street, N.W., Suite 800				
City	Washington	*State*	D.C.	*Zip Code*	20005
Country	U.S.A.	*Telephone*	202-783-6040	*Fax*	202-783-6031